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Influence of Parity and Calving Season on Resumption of Ovarian Cyclicity in Post-partum Murrah Buffaloes

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ABSTRACT

This study was designed to deduce the effect of parity and calving season on the resumption of ovarian cyclicity in post-partum Murrah buffaloes. For this study, 102 Murrah buffaloes (primiparous: 21; pluriparous: 81) calved between August 2015 and February 2016 were examined by transrectal ultrasonography. The first group of buffaloes between 26-days post-partum were taken and thereafter animals calved every next 10 days were grouped and added. Animals thus progressed and adjudged in 26-35, 36-45, 46-55, 56-65, 66-75, 76-85 and 86-95 days post-partum with an average of 30, 40, 50, 60, 70, 80 and 90 days post-partum, respectively. The day of second examination at 10 days interval of first group of animals also had first examination of second group and the same was followed for all study groups till cyclicity was confirmed in all study animals. In this investigation, 40.20, 65.69 and 81.37 % buffaloes resumed cyclicity within 30, 60 and 90 days post-partum, respectively. Significant ($p < 0.0001$) difference in resumption of cyclicity between primiparous and pluriparous buffaloes was observed. Onset of cyclicity in pluriparous buffaloes (37.24 ± 1.83 days post-partum) differed significantly ($p = 0.0001$) than primiparous (59.61 ± 6.10 days post-partum). Also, the onset of cyclicity was evident ($p < 0.0001$) during pre-breeding ($n = 60$) and resumption of ovarian cyclicity showed a significant trend ($p < 0.0001$) in relation to days post-partum. In summary, resumption of ovarian cyclicity differs significantly with parity and season of calving in post-partum Murrah buffaloes.

Key words: Buffalo, Murrah, Parity, Calving, Ovarian Cyclicity, Post-partum.

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INTRODUCTION

Resumption of ovarian cyclicity is an important factor to reduce the post-partum anestrus and improve the reproductive performance in buffaloes (El Wishy, 2007; Ahmed *et al.*, 2010). Resumption of post-partum activity and subsequent conception is affected by several factors *viz.* breed, nutrition plan, milk yield, suckling, uterine involution and calving season (Barile, 2005). Onset of post-partum ovarian cyclicity was determined by presence of corpus luteum (CL) (Sharma *et al.*, 2008). The incidence of post-partum anestrus in buffalo herds can range from 20 to 80%, with the greatest incidence during hot summers (Nanda *et al.*, 2003). Correct diagnosis of a CL and evaluating its functional status are important for the successful reproductive management of dairy animals, in addition to peripheral progesterone concentration (Perera, 2011). Mean interval to formation of first CL post calving was 23.8 ± 1.7 days, but only 52% of these CL were palpable in Nili-Ravi (Usmani *et al.*, 1985). Barman *et al.* (2011) reported 40 % buffaloes resuming cyclicity within 30 days and 80% of the buffaloes resuming cyclicity and 20 % remain acyclic till 90 days post-partum.

Similarly, Gokuldas *et al.* (2010) observed the resumption of ovarian cyclicity in Murrah buffaloes were 56.0 ± 2.37 days post-partum. Prolonged interval between parturition and resumption of cyclicity cause poor reproductive efficiency resulting in longer calving interval and thus causing great economic losses in the buffalo farm (El Wishy, 2007). Successful reproductive performance of buffalo depends upon uterine involution and early resumption of post-partum ovarian cyclicity (Perera, 2011). The present study was designed to observe the effect of parity and season on the resumption of ovarian cyclicity in post-partum Murrah buffaloes.

MATERIALS AND METHODS

Experimental animals and its management: The present study was conducted on 102 post-partum suckling buffaloes; primiparous (n = 21) and pluriparous (n = 81), calved between August 2015 and February 2016 at ICAR-Central Institute for Research on Buffaloes, Hisar. All the animals were healthy, calved normally and free from genital diseases. All the buffaloes were housed in concrete floor covered sheds with asbestos roofing and sides partly closed especially during milking. Thereafter, the animals were shifted in the open paddocks attached with each shed throughout the day with the provision of feeding and water troughs. The animals were confined in the covered shed during night hours in extreme winter season. The animals

were fed adequate quantity of green fodder (berseem, mustard, maize, sorghum) and dry fodder (wheat straw). Fresh and clean drinking water was provided free of choice. The concentrate supplemented with mineral mixture and common salt prepared by the Institute was fed individually to the animals before each milking. Feed requirement of buffaloes was adjusted keeping in view the milk yield. Animals were hand milked twice daily and suckling was allowed prior to each milking for letdown of milk.

Trans-rectal ultrasound examination: Ovarian ultrasound examination was carried out with a B-mode ultrasound scanner (Just Vision 200, Model 320A, Toshiba, Japan) equipped with a convex array multi-frequency intraoperative transducer at 7.5 MHz. A transrectal ultrasound scanning was made at 10 days interval starting from average 30 days (26 -35 days) post-partum to know the number and size of ovarian follicles and presence of CL. If a CL was detected by ultrasonography, the day of ovulation was calculated based on size of the CL, follicle and uterine tonicity as done earlier (Sharma *et al.*, 2012; Choudhary *et al.*, 2017; 2018). Ultrasound examination of each animal continued until detection of CL into one of the ovaries. Once CL was detected in either of the ovary, the animal was examined on next 10th day for the confirmation. When CL was detected on both the examination at 10 days interval the animal was confirmed cyclic and was excluded for further examination. Ultrasound examination was carried and once animal was confirmed cyclic by the presence of CL on two consecutive ultrasound examinations, it was not brought for further examination. Monitoring of onset of ovarian cyclicity was carried during the pre-breeding (August-October) and breeding season (November to February).

Blood sampling, progesterone assay : Progesterone assay was carried out to confirm the cyclicity of animals as animals with serum progesterone more than 1 ng/mL were considered cyclic. For this, blood sample (10 ml) were collected via jugular venipuncture in serum clot activated vacutainer (VACUETTE®) serum tube. First blood sample from each buffalo was taken when corpus luteum (CL) was found at first instance by ultrasound examination and second sample at 10 days after, for progesterone assay. Those buffaloes which remain acyclic till 80 days, samples were taken at 80- and 90-days post-partum for progesterone assay. The blood samples collected during this period were chilled on ice, transported to the laboratory and centrifuged at 3000 rpm for 15 minutes at 4°C and serum was harvested and stored in duplicate each in 1.5 mL tube at -20°C till it is assayed for progesterone. Serum samples were thawed at 4° C, 12 hr before assay. A solid phase enzyme immunoassay kit of (XEMA Co., Ltd, Moscow, Russia) was

used for the quantitative determination of progesterone in serum samples. The intra- and inter-assay variations were < 5 and < 10 , respectively having a sensitivity of 0.15 ng/mL.

Statistical analysis: Data was analyzed with SPSS (Version 16) using Chi-Square test to find the difference in the resumption of cyclicity status in study animals based on parity, season and days post-partum under study. Results were considered significant at $p < 0.05$.

RESULTS AND DISCUSSION

Resumption of ovarian cyclicity is a key factor to reduce the post-partum anestrus and improving reproductive performance in buffaloes (Khasatiya *et al.*, 2006; El Wishy, 2007a and Ahmed *et al.*, 2010). Prolonged interval between parturition and resumption of cyclicity causes poor reproductive efficiency resulting in longer calving interval and greater economic losses in any buffalo herd (ElWishy, 2007; Perera, 2011). Successful reproduction of buffalo depends upon uterine involution and early resumption of post-partum ovarian cyclicity (Perera, 2011). Our results are concordant with earlier study in buffaloes (Abdalla, 2003), wherein the first ovulation observed was at 29.2 ± 1.2 days after calving and 88% buffaloes resumed cyclicity till 90 days post-partum, but Yindee *et al.* (2007) found an interval of 39.81 ± 3.38 days from calving to first ovulation in buffaloes. Similar results (38.5 ± 3.9 days post-partum) was reported by Barkawi *et al.* (1998) in Egyptian buffaloes and Rijasnaz *et al.* (2012) reported post-partum interval to resumption of ovarian cyclicity was 27.8 ± 1.81 days in suckling Murrah buffaloes. In our study, significant ($p < 0.0001$) difference in resumption of cyclicity between primiparous and pluriparous buffaloes was observed, which revealed that 61.90% (13/21) primiparous buffaloes became cyclic within 59.61 ± 6.10 days post-partum. Resumption of post-partum ovarian cyclicity was earlier ($p < 0.05$) (37.24 ± 1.83 days post-partum) in pluriparous than primiparous buffaloes (59.61 ± 6.10 days post-partum). It was observed that percentage of cyclic animals was seen more in pluriparous (86.41%) than primiparous (68.42%) buffaloes. In pluriparous buffaloes, 86.41

% (70/81) resume cyclicity. Onset of cyclicity in pluriparous buffaloes (37.24 ± 1.83 days post-partum) differ significantly ($p = 0.0001$) than primiparous (59.61 ± 6.10 days post-partum) (Table 1). Earlier workers (Ali and El-Sheikh 1983; Devaraj and Janakiraman, 1986) reported the resumption of cyclicity in primiparous buffaloes was longer than pluriparous buffaloes. Meikle *et al.* (2004) also found that initiation of ovarian cyclicity was delayed in primiparous than multiparous cows. But, no significant differences between parity was reported earlier (El-Wishy 2007). In Our study, serum progesterone of cyclic ($n=83$) and acyclic buffaloes ($n=19$) were >1 and <1 ng/ml respectively ($p < 0.05$). Yotov and Antanasov (2013) reported that faster development of ovulatory follicles followed by ovulation and CL formation in multiparous than in primiparous Bulgarian Murrah buffaloes. Interval between parturition and first identification of CL was 38.6 ± 12.46 days and 31.0 ± 12.10 days in primiparous and multiparous respectively, indicated an earlier resumption of the ovarian activity after calving in multiparous than primiparous. Our study also revealed that 54% buffaloes ovulated before day 40 post-partum. This is significant as it allowed the animals to experience one or two estrous cycles for successful breeding before 70 to 90 days post-partum, by which buffaloes must be pregnant to maintain a calving interval of 13–14 months (El-Wishy, 2007).

With respect to season wise distribution of calving, the maximum number of calving occurred during pre-breeding ($n = 64$). Maximum number of calving's occurred in the month of pre-breeding season ($n = 64$) and as compared to breeding season ($n = 38$), however, the onset of cyclicity was more ($p < 0.0001$) during pre-breeding ($n = 60$) than breeding ($n = 23$) season (Table 2). The results of onset of cyclicity indicated that significant ($p < 0.0001$) number of buffaloes (93.75%) calved during pre-breeding season resumed cyclicity. It is noteworthy that significant number of buffaloes exhibit estrus during the of short-day length period than during the period of long day length (Tailor *et al.*, 1990). Also, studies have shown that buffaloes calving during rainy and monsoon seasons had shorter anestrus periods than calving in other seasons (Tailor *et al.*, 1997; Sule *et al.*, 2001).

Table 1: Resumption of cyclicity status based on parity in Murrah Buffaloes.

Parity	Cyclic				Acyclic			
	<i>n</i>	Chi-square; P value	Mean \pm SE	P value	<i>n</i>	Chi-square; P value	Mean \pm SE	P value
Primiparous	13	75.566; < 0.0001	59.61 ± 6.10	0.0001	8	0.421; 0.5164	92.00 ± 0.94	0.39
Pluriparous	70		37.24 ± 1.83		11		90.72 ± 1.03	

Table 2: Resumption of cyclicity status in Murrah buffaloes calved in different season.

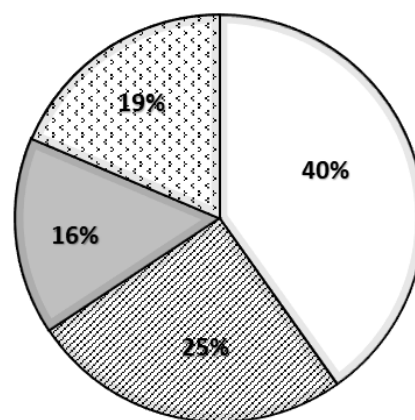
Season	Calving (n)	Resumed cyclicity (n)	Cyclicity (%)	Chi-square; P value
Pre-breeding	64	60	93.75	31.229;
Breeding	38	23	60.52	< 0.0001
Total	102	83	81.37	

Onset of post-partum ovarian cyclicity was determined by presence of CL (Sharma *et al.*, 2008) as well as peripheral progesterone concentration (Choudhary *et al.*, 2018). Correct diagnosis of a CL and estimating its functional status are important for the successful reproductive management of dairy animals. The present study showed that 40.27, 13.73, 8.82, 2.94, 7.84, 2.94 and 4.90 % of buffaloes resumed cyclicity at 30, 40, 50, 60, 70, 80- and 90-days post-partum period. In first month 40.72 % (n = 41) buffaloes resumed cyclicity in 27.36 ± 0.44 days post-partum. Subsequently, 27.45 % (n = 28) buffaloes became cyclic in 43.25 ± 1.63 days post-partum (31-60 days) and 13.72 % (n = 14) buffaloes resumed cyclicity in 74.78 ± 2.85 days post-partum (61-90 d). Out of 102 buffaloes 18.62% (n = 19) remained acyclic till 90 days post-partum (Table 3; Fig.1). The post-partum interval to resumption of ovarian cyclicity was 27.8±1.81 days in sucking Murrah buffaloes (Rijasnaz *et al.*, 2012). Barman *et al.* (2011) reported that first post-partum ovulation occurred at 27.85±4.06 days post-partum and 40% buffaloes resuming cyclicity within 30 days and 80% of the buffaloes resuming cyclicity and 20% remain acyclic till 90 days post-partum. Our peripheral progesterone estimates corroborate with the previous findings (Perera, 2011; Choudhary *et al.*, 2017; 2018). Resumption of post-partum activity and subsequent conception is affected by several factors such as breed, nutrition, milk yield, suckling, uterine involution and season of calving (Barile, 2005; Perera, 2011). In sub-tropical zone and at higher altitude, buffaloes exhibiting estrus during the period of short-day length is significantly greater than during the long day length which shows that decreasing day light is a stronger determinant of resumption of post-partum ovarian activity. Reproductive period is longer near equator where the light /dark ratio is constant throughout the year (Campanile *et al.*, 2010; Zicarelli, 1997). Also, our results agreed with Gokuldas *et al.* (2010) who reported the resumption of ovarian cyclicity in Murrah buffaloes were 56.0±2.37 days post-partum and first ovulation occurred at 57.0±2.37 days post-partum, however proportion of animals resuming cyclicity by days 60 and days 90 post-partum were 38.46% and 61.54% respectively, with 50% incidence of silent estrus. Usmani *et al.* (1984) reported that in 53 buffaloes the interval from calving to 1st estrus averaged 56.4±3.9 days, with a range of 18-154 days.

Table 3: Resumption of ovarian cyclicity at different days post-partum in Murrah Buffaloes (n = 102).

Days post-partum	No. of animals (%)	Cumulative (%)	Resumption of cyclicity (post-partum days) (Mean ± SE)	Chi-square; P value
30	41 (40.20)	40.20	27.37 ± 0.44	
40	14 (13.73)	53.93	35.00 ± 0.80	
50	9 (8.82)	62.75	46.00 ± 0.75	233.5;
60	3 (2.94)	65.69	52.33 ± 1.88	< 0.0001
70	8 (7.84)	73.53	63.33 ± 0.73	
80	3 (2.94)	76.47	77.00 ± 1.53	
90	5 (4.90)	81.37	86.40 ± 1.81	

□ <30 Days ▨ 31-60 Days ■ 61-90 Days ◻ Acyclic

**Fig.1:** Post-partum resumption of cyclicity (n = 102)

CONCLUSIONS

This study reports that season of calving and parity influenced the resumption of ovarian cyclicity in post-partum buffaloes and majority of buffaloes (> 40 %) resumed cyclicity within 30 days post-partum.

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CONFLICT OF INTEREST

None.

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